(a) placing a biological sample comprising said nucleic acid sequence in a capillary vessel;

(b) raising the temperature of the biological sample from a first temperature to a second temperature wherein the second temperature is at least 15°C higher than the first temperature;

(c) holding the biological sample at the second temperature for a predetermined amount of time;

(d) lowering the temperature of the biological sample from the second temperature to at least the first temperature;

(e) holding the biological sample at a temperature at least as low as the first temperature for a pre-determined length of time;

(f) raising the temperature of the biological sample to the second temperature; and

(g) repeating steps c through f, wherein those steps are completed in approximately 60 seconds or less.

31. The improved method of claim 30 wherein the capillary vessel defines a volume ranging from about 10 microliters to about 100 microliters.

32. The improved method of claim 30 wherein the pre-determined length of time for holding step (c) is less than one second.

33. The improved method of claim 30 wherein step (d) comprises lowering the temperature of the biological sample to a third temperature that is below the first temperature, step (e) comprises holding the biological sample at the third temperature for a pre-determined length of time, said method further comprising the step of raising the temperature of the biological sample back to the first temperature and holding the sample at the first temperature for a pre-determined length of time before proceeding to step (f).

34. The improved method of claim 33 wherein the pre-determined length of time for holding step (e) is less than one second.

35. An improved method of modifying a biological sample by subjecting the biological sample to multiple cycles of controlled rapid heating and cooling, said method comprising the steps of placing the sample in a capillary vessel and thermally cycling the

sample by contacting the capillary vessel with a heated fluid to raise the temperature of the sample from a first temperature to a second temperature, cooling the sample from said second temperature to a temperature at least as low as the first temperature, and heating the sample back to the second temperature; wherein the difference between the first temperature and the second temperature is at least 15°C, and wherein the heating and cooling step are completed in approximately 60 seconds or less, while the temperature homogeneity in the sample is maintained within plus or minus 1°C during the heating and cooling steps.

36. In a method of amplifying a DNA sequence by thermal cycling of the nucleic acid sequence in the presence of a thermostable DNA polymerase, wherein each cycle comprises the steps of heating a biological sample containing said DNA sequence to a denaturing temperature; holding the biological sample at the denaturing temperature; cooling the sample to an annealing temperature, holding the temperature at the annealing temperature for a pre-determined amount of time; warming the sample to an elongation temperature; and holding the sample at the elongation temperature for a predetermined amount of time; the improvement

comprising limiting the holding time of the annealing and denaturing steps to less than one second each.

37. A method for improving the purity of a product produced by polymerase chain reaction wherein a DNA sequence is amplified by thermal cycling of the sequence in an aqueous sample in the presence of a thermostable polymerase and wherein each thermal cycle comprises heating the aqueous sample to a denaturation temperature, holding it at the denaturation temperature for a predetermined period of time, cooling the sample to an annealing temperature and holding it at the annealing temperature for a predetermined period of time, the method comprising the step of limiting the time the sample is held at the annealing temperature to less than 5 seconds.

38. The method of claim 37 wherein the sample is held at the annealing temperature for less than one second.

39. The method of claim 37 further comprising the step of cooling the sample from the denaturation temperature to the annealing temperature at a rate of at least about 1.48°C per second.

40. The method of claim 37 wherein each thermal cycle is completed in less than 60 seconds.

41. The method of claim 37 wherein the period of time the sample is held at the denaturation temperature during each thermal cycle is less than 32 seconds.

42. The method of claim 41 wherein the period of time the sample is held at the denaturation temperature during each thermal cycle is less than one second.

43. A method for increasing the yield of an amplified DNA sequence by use of polymerase chain reaction wherein the DNA sequence is thermally cycled in an aqueous sample in the presence of a thermostable polymerase and wherein during each thermal cycle the sample is heated to a denaturation temperature and held for a predetermined period of time, said method comprising the step of limiting the time the sample is held at the denaturation temperature to less than 32 seconds.

44. The method of claim 43 wherein the sample is held at the denaturation temperature for less than one second.

45. The method of claim 43 wherein each thermal cycle is completed in less than approximately 60 seconds.

46. In the construction of a thermal cycling device suitable for performing polymerase chain reaction DNA amplification in which the device comprises a closed-loop system, a fan for moving air in the closed-loop system to transfer heat from a heat source to a sample compartment in said closed-loop system, a temperature sensor in the sample compartment, and a programmable temperature controller for controlling the temperature in the sample compartment, the improvement which comprises minimizing the thermal mass of the device components in the closed-loop system in contact with the air, installing air mixing baffles in the closed-loop system, and selecting the fan and the heat source so that the temperature in the sample compartment can be cycled between a DNA denaturation temperature of at least 90° and a DNA-annealing temperature in the range of about 50° to about 55°C in approximately 30 to approximately 60 seconds while maintaining a temperature gradient in the sample compartment of less than 2°C.

47. The improvement of claim 46 wherein the fan has the capacity of at least 75 cubic feet of air per minute.

48. A thermal cycling device for optimizing yield and purity of amplified DNA prepared by a process comprising thermal cycling of an aqueous sample comprising a DNA sequence and a thermostable polymerase between a DNA denaturation temperature and a lower DNA annealing temperature, said device comprising

a chamber for substantially confining and directing flow of air contained therein along a closed-loop path,

a sample compartment located in said chamber within said closed-loop path, a fan for moving air along said closed-loop path, means for heating air in said closed-loop path,

means for mixing air in the closed-loop path between the air heating means and the sample compartment,

a thermal sensor for providing a signal related to the temperature of said sample compartment,

a vent in the chamber to direct air from the closed-loop path and means for opening and closing said vent, and

a programmable controller for receiving the signal from the thermal sensor and for controlling the air-heating means and the vent opening and closing means to cycle the temperature of air in the sample compartment wherein the controller is programmable to cycle the temperature in the sample compartment between a DNA-denaturation temperature and a DNA-annealing temperature in 60

seconds or less while maintaining a temperature gradient in the sample compartment of less than 10°C.

49. The device of claim 48 wherein during thermal cycling, the temperature gradient in the sample compartment is less than 2°C.

50. The device of claim 48 wherein during thermal cycling the temperature of a sample in the sample compartment can be held at a DNA-denaturation temperature or a DNA-annealing temperature for less than 1 second.

51. The device of claim 48 wherein the temperature in the sample chamber can be cycled between a DNA-denaturation temperature and a DNA-annealing temperature in approximately 30 seconds.

52. The device of claim 51 wherein during thermal cycling, the temperature gradient in the sample compartment is less than 2°C.

53. The device of claim 48 constructed so that the thermal mass of the device components in contact with air in the closed-loop path is minimized.

54. The device of claim 48 wherein in the capacity of the fan is at least 75 ft.3/min.

55. The device of claim 48 wherein the temperature of a sample in the sample compartment can be uniformly cycled at a rate of temperature change of at least 1.48°C/sec.

56. The device of claim 48 wherein the temperature of a sample in the sample compartment can he uniformly cycled at a rate of temperature change of at least 4.11°C/sec.

REMARKS

After entry of this amendment, it is believed that all of the claims now pending are allowable. Thus, favorable action concerning these claims is respectfully requested. If any impediment to the allowance of these claims remains after entry of this Amendment and consideration of these remarks the Examiner is invited to initiate a telephone interview with the undersigned.